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Sequential Age Dependent Gross Morphological Changes in the Spleen and Harderian Gland of Turkeys

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ABSTRACT: The study on sequential age dependent changes of spleen and harderian gland in broad breasted bronze turkeys was aimed to understand the immunological role of these secondary lymphoid organs in this species according to age. To explore the age related changes in its gross morphology, the samples were taken from day old to ten months of age at monthly intervals. There were no changes noticed in the shape and morphological features of both organs which maintained its elliptical shape and human foot print shape respectively throughout the study. The variation was existed only in its morphometric parameters such as weight, length and width. The weight of both organs showed steady increase till six months of age and thereafter it was slowly reduced. The length and width of the spleen were also steadily increased along with splenic weight and showed decline phase after six months of age. There was no significant difference in the weight observed between left and right Harderian gland. The length and width of the harderian gland also showed decreasing trend along with weight as age advanced.

Keywords: Age dependent changes, Spleen, Harderian gland, Broad Breasted Bronze turkey.

INTRODUCTION

The immune system of birds plays a vital role in the maintenance of health by cell mediated and humoral immunity (Penchev, 2020). The functional immune cells leave from the primary lymphoid organs, thymus and bursa of Fabricius and accumulate in secondary lymphoid organs such as spleen and harderian gland. The spleen is the important secondary immune organ which contains both B and T cells and has significant role to resist antigens (John, 1994). The Harderian gland is a bursa dependent organ described as a head associated lymphatic tissue and populated by interstitial B lymphocytes, plasma cells and few T cells between the exocrine portions which function in lubrication of eye (Maslak & Reynolds 1995). The major accumulating lymphatic cells of Harderian gland are B cells and are converted into plasma cells which play a role in humoral antibody response against infection.

As the primary immune organs regress after sexual maturity, the immunity is mainly conferred by secondary lymphoid tissues in post- pubertal age. This study was formulated with the aim to reveal the gross morphological changes occur in the secondary lymphoid tissues such as spleen and harderian gland in both pre and post sexual maturity period.

MATERIALS AND METHODS

The post-natal immune organs, the spleen and Harderian gland specimens were collected at the time of Jayachitra & Sivagnanam Biological Forum – An International Journal 15(5): 1435-1438(2023)

slaughter in broad breasted turkeys. They were collected at monthly intervals from day-old to ten months of age. Each group was consisted of six birds and was apparently healthy and vaccinated against Newcastle disease. The body cavity of birds was opened by through a mid-ventral incision behind the keel region to collect the spleen. A sagittal section of head was done to locate the Harderian gland and was removed from the orbital cavity. After collection, they were washed in normal saline, weighed and utilized to take morphometrical parameters. The morphological features and age dependent changes of each organ were studied at monthly intervals. The morphometrical values were analyzed statistically by two way ANOVA followed by Duncan's test to reveal the significant age related changes.

RESULTS AND DISCUSSION

A. Spleen

Grossly, the age related changes were not noticed in the shape and morphological features of spleen which maintained its elliptical shape throughout the study but the variation was noticed only in morphometric parameters such as weight, length and width (Table 1). The weight of the spleen showed steady increase and reached peak weight of 4.02 ± 0.02 gm in tom and 2.63 ± 0.02 gm in hen turkeys at six months of age. Thereafter it was slowly reduced and achieved the weight of 2.20 ± 0.02 gm in tom and 1.20 ± 0.02 gm in hen

at ten months of age (Plate 1). In accordance, the splenic weight was increased gradually from first to eleventh week and tends to decrease from 13th week in duck (Hashimoto & Sugimura 1977) and the relative weight of spleen was reached maximum at 5 weeks of age and then reduced afterwards in guinea fowl (Surjith et al., 2007) whereas Liman & Bayram (2011) stated that the weight of spleen was increased from day old to 60 days of age in quail. The sex hormones played a major role in growth of spleen and caused no reduction in weight in castrated birds but, the drop in weight was noticed in non-castrated birds after sexual maturity in Japanese quail (Mase & Oishi 1991). John (1994) also stated that an inverse relationship was present between gonad and spleen size. The estradiol and testosterone treatment in pigeon caused the lymphocyte destruction and reduction in white pulp of spleen.

In contrast, Piao *et al.* (2018) in Chinese yellow quail stated that the splenic weight was increased from dayold to 14 weeks of age and no significant difference was observed from 14^{th} to 38^{th} week of age. Tarek *et al.* (2018) in broiler chicken described the growth of spleen in two phases. First phase showed gradual increase in weight from first week (0.1gm) to seventh week (3.7gm) and the second stationary phase started from eighth week onwards.

The length and width of the spleen was steadily increased along with splenic weight and showed decline phase after six months of age (Table 1). This observation was agreeing with Indu et al. (2011) in White Pekin ducks who stated that the splenic weight, length and width were increased gradually from day-old $(0.01\text{gm} \times 0.4\text{cm} \times 0.30\text{cm})$ and reached the maximum at 110 days of age (1.1gm \times 2.50cm \times 1.60cm). Afterwards it showed the reduction in its morphometrical parameters and recorded as $0.74 \text{gm} \times$ 1.74 cm \times 1.24 cm at 155 days of age. In contrary, Verma et al. (2018) in Kadaknath fowl recorded the average length and width as 7.35 ± 1.36 mm \times 4.78±0.53mm. 15.1±1.25mm 9.5±1.33mm, X 21.1 \pm 0.34mm × 16.03 \pm 0.507mm, 22.45 \pm 0.32mm × 17.08±0.45mm in 1-8, 9-16, 17-24, 25-32 weeks of age respectively.

B. Harderian gland

Harderian gland also not showed the age dependent changes in its gross morphological features. Throughout the study period it was human foot print in shape (Plate 2). In both the sexes, the Harderian gland attained its maximum weight at sixth month of age and thereafter slightly reduced as age advanced. The weight of Harderian gland was recorded as 0.214 ± 0.01 gm in tom and 0.147 ± 0.01 gm in hen at six months and 0.175 ± 0.01 gm in tom and 0.125 ± 0.01 gm in hen at 10 months of age. The length and width of the gland also increased along with weight and attained its maximum at six months. Afterwards, they showed decreasing trend along with weight as age advanced (Table 2).

Similarly, the length and width of Harderian gland was higher in male and recorded as $6.00\pm0.41 \times 3.13\pm0.31$ mm (Male), $5.38\pm0.24 \times 2.63\pm0.24$ mm (Female) and $7.38\pm0.23 \times 4.38\pm0.24$ mm (Male),

 $6.50\pm0.29 \times 3.8\pm0.21$ mm (Female) in native and broiler chicken respectively at six months of age (Jahan *et al.*, 2006). Nawrot *et al.* (2016) in capercaillie also reported that the weight of organ was higher in adult male than female and measured the length and width as $12.17\pm0.63 \times 5.64\pm0.21$ mm in male and $10.21\pm0.54 \times$ 4.08 ± 0.16 mm in female. Ashok *et al.* (2000) documented the length and width of Harderian gland as 17.66mm and 6.2 mm respectively in adult White leghorn chicken.

In contrary, Dimitrov (2012) in common bronze turkey recorded the weight, length and width of Harderian gland as 229.92 ± 0.14 mg $\times 24.60\pm0.57$ mm \times 8.05 ± 0.28 mm at eight months of age. This variation might be attributed to difference in variety and its geographical location of farming condition. The slight decrease in the morphometric parameters of Harderian gland after six months of age might be correlated with reduction in the weight of bursa of Fabricius as it was bursa dependent organ to receive B lymphocytes.

In accordance with reports made by Frahmand & Mohammadpour (2015) in Canadian ostrich, there was no significant difference observed between weight, length and width of left and right gland. Due to the limitations on age and sex dependent studies on Harderian gland from day-old to ten months of age, the comparison could not be made.



Plate 1. Photograph showing the development of spleen from one month to ten months of age in broad breasted bronze turkey.



Plate 2. Photograph showing the development of harderian gland from one month to ten months of age in broad breasted bronze turkey.

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	Weight (gm)		Length (mm)		Width (mm)	
Age	Male	Female	Male	Female	Male	Female
Day-old	0.02±0.004 ^{bA}	0.01±0.006 ^{aA}	4.50±0.050 ^{bA}	4.00±0.050 ^{aA}	2.33±0.061 ^{bA}	1.90±0.049 ^{aA}
1 month	0.26 ± 0.036^{bB}	0.13±0.007 ^{aA}	9.30±0.049 ^{bB}	8.73±0.061 ^{aB}	5.07±0.061 ^{bB}	4.80±0.049 ^{aB}
2 months	1.10±0.019 ^{bC}	0.85±0.023 ^{aB}	14.07±0.061 ^{bC}	12.57±0.072 ^{aC}	9.90±0.050 ^{bD}	8.57±0.101 ^{aC}
3 months	1.62±0.022 ^{bD}	1.06±0.025 ^{aC}	18.33±0.079 ^{bD}	15.43±0.087 ^{aD}	10.27±0.072 ^{bE}	9.53±0.079 ^{aD}
4 months	2.56±0.059 ^{bF}	1.24±0.030 ^{aD}	22.10±0.049 ^{bF}	17.03±0.061 ^{aF}	11.10±0.070 ^{bF}	10.40 ± 0.071^{aD}
5 months	3.28±0.028 ^{bG}	1.96±0.034 ^{aG}	25.10±0.070 ^{bI}	20.07±0.061aG	13.10±0.070 ^{bH}	11.37±0.061 ^{aE}
6 months	4.02±0.004 ^{bI}	2.63±0.038 ^{aI}	27.20±0.049 ^{bJ}	24.57±0.079 ^{aI}	14.00±0.049 ^{bJ}	12.00±0.050 ^{aF}
7 months	3.79±0.019 ^{bH}	2.31±0.027 ^{aH}	25.00±0.049 ^{bI}	21.20±0.049 ^{aH}	13.57±0.061 ^{bI}	11.50±0.086 ^{aE}
8 months	3.21±0.070 ^{bG}	1.79±0.012 ^{aF}	24.07±0.061 ^{bH}	18.50±0.086 ^{aF}	12.10±0.070 ^{bG}	10.10±0.099 ^{aD}
9 months	2.32±0.030 ^{bE}	1.39±0.031 ^{aE}	22.67±0.079bG	16.47 ± 0.078^{aE}	10.07±0.070 ^{bD}	9.13±0.062 ^{aD}
10 months	2.20±0.031 ^{bD}	1.20±0.050 ^{aCD}	21.27±0.077 ^{bE}	15.33±0.071 ^{aD}	8.17±0.079 ^{bC}	9.07±0.071 ^{aD}

Table 1: Mean±SE of morphometric measurements of spleen in turkeys.

Mean±SE with same superscript between columns (ab..), rows (AB..) do not differ significantly (P<0.05)

Table 2: Mean±SE of morphometric measurements of Harderian gland in turkeys.

Age	Weight (gm)		Length (mm)		Width (mm)	
	Male	Female	Male	Female	Male	Female
Day-old	0.012±0.005 ^{bA}	0.009 ± 0.005^{aA}	8.80±0.049 ^{bA}	8.03±0.061 ^{aA}	1.60±0.019 ^{bA}	1.36±0.034 ^{aA}
1 month	0.032 ± 0.005^{bB}	$0.029{\pm}0.008^{aB}$	10.80±0.053 ^{bB}	10.07±0.061 ^{aB}	2.54±0.035 ^{bB}	2.34±0.022 ^{aB}
2 months	0.076±0.004 ^{bC}	0.054 ± 0.006^{aC}	14.07±0.061 ^{bC}	13.63±0.061 ^{aC}	3.80±0.049 ^{bC}	3.36±0.029 ^{aC}
3 months	0.162±0.004 ^{bD}	0.102 ± 0.008^{aD}	16.30±0.070 ^{bD}	14.80±0.049 ^{aD}	3.90±0.049 ^{bC}	3.63±0.033 ^{aD}
4 months	0.173±0.006 ^{bE}	0.125 ± 0.005^{aE}	17.27±0.074 ^{bE}	15.47±0.061 ^{aE}	4.43±0.022 ^{bF}	4.18±0.022 ^{aFG}
5 months	0.185±0.009 ^{bG}	0.136±0.005 ^{aG}	19.50±0.049 ^{bFG}	17.90 ± 0.049^{aH}	4.50±0.049 ^{bF}	4.30±0.070 ^{aG}
6 months	0.214±0.009 ^{bI}	0.147 ± 0.010^{aI}	21.37±0.071 ^{bJ}	19.27±0.072 ^{aJ}	4.47±0.061 ^{bF}	4.43±0.037 ^{aH}
7 months	0.193±0.006 ^{bH}	0.139±0.006 ^{aH}	20.77±0.061 ^{bI}	18.77 ± 0.061^{aI}	4.37±0.071 ^{bEF}	4.23±0.067 ^{aG}
8 months	0.187±0.007 ^{bG}	0.136±0.010 ^{aG}	19.67±0.061 ^{bH}	17.70±0.070 ^{aG}	4.27±0.071 ^{bBE}	4.23±0.070 ^{aF}
9 months	0.179±0.008 ^{bF}	0.131 ± 0.008^{aF}	19.53±0.071 ^{bGH}	17.47±0.079 ^{aF}	4.23±0.061 ^{bDE}	4.10±0.061 ^{aE}
10 months	0.175±0.008 ^{bE}	0.125 ± 0.005^{aE}	19.37±0.037 ^{bF}	17.37±0.053 ^{aF}	4.13±0.037 ^{bD}	3.93±0.049 ^{aE}

Mean±SE with same superscript between columns (ab..), rows (AB..) do not differ significantly (P<0.05)

CONCLUSIONS

This study on post hatch age dependent morphological evaluation of spleen and harderian gland of turkey revealed that the correlation exist between the growth of these organs and sexual maturity of turkeys. Upon sexual maturity, they showed decline phase on its morphological parameters in both the sexes. It might be due to influence of sexual hormones and regression of primary immune organs which play a role in populating the lymphocytes to secondary lymphoid tissues.

FUTURE SCOPE

Further study on morphological and bio-metrical evaluation of interrelationship between secondary lymphoid tissues such as spleen and harderian gland with primary lymphoid organs and gonads will be initiated.

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